

AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph beginning on page 26, line 31 with the following rewritten paragraph:

This invention also contemplates the use of derivatives of 312C2s other than variations in amino acid sequence or glycosylation. Such derivatives may involve covalent or aggregative association with chemical moieties. Covalent or aggregative derivatives will be useful as immunogens, as reagents in immunoassays, or in purification methods such as for affinity purification of binding partners, e.g., other antigens. A 312C2 can be immobilized by covalent bonding to a solid support such as cyanogen bromide-activated ~~SEPHAROSE~~ SEPHAROSE®, a bead-formed agarose gel filtration matrix, by methods which are well known in the art, or adsorbed onto polyolefin surfaces, with or without glutaraldehyde cross-linking, for use in the assay or purification of anti-312C2 antibodies or an alternative binding composition. Western blot techniques are also common. The 312C2s can also be labeled with a detectable group, e.g., for use in diagnostic assays. Purification of 312C2 may be effected by an immobilized antibody or complementary binding partner.

Please replace the paragraph beginning on page 56, line 21 with the following rewritten paragraph:

The PCR-based subtraction system developed by Wang and Brown (1991) Proc. Natl. Acad. Sci. USA 88:11505-11509, was modified to apply to plasmid cDNA libraries. A cDNA library specific for activated $\alpha\beta$ DN thymocytes was generated using 100 μ g of the unstimulated $\alpha\beta$ DN cDNA library DNA digested with XbaI, NotI, and ScaI as driver DNA and 5 μ g of the stimulated $\alpha\beta$ DN cDNA library DNA as tracer DNA. Following restriction digestion, the driver DNA was treated with DNA polymerase Klenow fragment to fill-in the restriction sites. After ethanol precipitation, the DNA was dissolved in 100 μ l of water, heat-denatured and mixed with 100 μ l (100 μ g) of ~~Photoprobe~~ PHOTOPROBE® biotin, a photoreactive molecule-biotin complex (Vector Laboratories, Burlingame, CA). The driver DNA was then irradiated with a 270-W sunlamp on ice for 20 min. 50 μ l more ~~Photoprobe~~ PHOTOPROBE® biotin, a

photoreactive molecule-biotin complex, was added and the biotinylation reaction was repeated. After butanol extraction, the photobiotinylated DNA (driver-U) was ethanol-precipitated and dissolved in 30 μ l of 10 mM Tris-HCl and 1 mM EDTA, pH 8 (TE). As tracer DNA, 5 μ g of stimulated $\alpha\beta$ DN cDNA was digested with XbaI and NotI; ethanol precipitated; and dissolved in 4 μ l of TE (tracer-S). Tracer-S was mixed with 15 μ l of driver-U, 1 μ l (10 μ g) of E. coli tRNA (Sigma, St. Louis, MO), and 20 μ l of 2 x hybridization buffer (1.5 M NaCl, 10 mM EDTA, 50 mM HEPES, pH 7.5, 0.2% SDS), overlaid with mineral oil, and heat-denatured. The sample tube was immediately transferred into a 68° C water bath and incubated for 20 h. The reaction mixture was then subjected to streptavidin treatment followed by phenol/chloroform extraction. Subtracted DNA was precipitated, dissolved in 12 μ l of TE, mixed with 8 μ l of driver-U and 20 μ l of 2 x hybridization buffer, and then incubated at 68° C for 2 h. After streptavidin treatment, the remaining DNA was ligated with 250 ng of a purified XbaI / NotI fragment of pJFE-14 and then transformed into electro-competent E. coli cells to generate the activation specific $\alpha\beta$ DN subtracted library (S1). 100 independent clones were randomly picked and screened by hybridization using a cocktail of known cytokine cDNAs. Plasmid DNA's were prepared from clones that did not hybridize to the cytokine probes. These clones were grouped by insert size and further characterized by DNA sequencing. Clones corresponding to the 312C2 were isolated.